Annals of Gastroenterology (2026) **39,** 1-6

Utilization and diagnostic yield of multitarget stool DNA testing in a large rural healthcare system

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Abstract

Background Colorectal cancer (CRC) ranks as the third most prevalent cancer and second leading cause of cancer-related deaths. The multitarget stool DNA (MT-sDNA) test is one of the screening modalities for CRC. We describe its use and its diagnostic yield among patients with a positive MT-sDNA test.

Methods This was a single-center retrospective study of patients with positive MT-sDNA test results, followed by colonoscopy between May 2023 and May 2024. Demographics and colonoscopy findings were collected from electronic medical records. The primary outcome was the proportion of positive MT-sDNA tests associated with advanced neoplasia (advanced adenomas or colorectal cancer) among patients who completed follow-up colonoscopy.

Results A total of 4669 patients had active MT-sDNA orders, of whom 3536 (75.7%) completed the test. A total of 408 patients with positive MT-sDNA results were included in the analysis (mean age 65.9±8.7 years). Off-label MT-sDNA was ordered in 111 (27.2%) patients. Among patients with positive MT-sDNA results, 275 (67.4%) underwent follow-up colonoscopy; complete pathology data were available for 270 (66.2%). Of these, 1 patient (0.4%) was diagnosed with CRC, and 35 (13.0%) had advanced adenomas.

Conclusions In a rural, community-based cohort, off-label use of MT-sDNA was common, and was associated with a high proportion of positive tests not associated with advanced neoplasia. Failure to complete follow-up colonoscopy after a positive test was more frequent among patients with tobacco and alcohol use disorders. These findings underscore the need for targeted provider education and patient-centered interventions to improve diagnostic follow up and healthcare resource utilization.

Keywords Multitarget stool DNA, colorectal cancer, colonoscopy

Ann Gastroenterol 2026; 39 (XX): 1-6

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Conflict of Interest: None

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Received 7 February 2026; accepted 18 July 2026; published online 17 September 2026

DOI: https://doi.org/10.20524/aog.2026.1108

Introduction

Colorectal cancer (CRC) ranks as the third most prevalent cancer globally and is the second leading cause of cancer-related deaths [1]. In the United States, 153,000 people are annually diagnosed with colorectal cancer [2]. Over recent decades, its incidence in high-income countries has either stabilized or declined [1]. The incidence and mortality of CRC rise significantly with age, with the median age of diagnosis nearing 70 years in developed nations [3]. In addition to age, key risk factors include male sex, smoking, heavy alcohol consumption, lack of physical activity, high intake of red and processed meats, being overweight, and a family history of CRC [3]. The occurrence of CRC in individuals aged under 50, known as "early-onset CRC", has been increasing [4]. Since randomized controlled trials assessing the benefits of lowering CRC screening age are lacking, current recommendations are derived mainly from decision-analytic models [4]. The decline in CRC incidence over the past decade among individuals aged

over 55 years is probably driven by a rise in screening rates, which increased from 38% in 2000 to 59% in 2013 for adults aged 50 and older [5].

The Food and Drug Administration (FDA) has approved multitarget stool DNA testing (MT-sDNA), known as the Cologuard® test, which is now recommended every 3 years for screening purposes [6]. While its convenience may improve screening adherence, inappropriate ordering outside the recommended indications can reduce its effectiveness, increase healthcare costs, and potentially delay appropriate diagnostic evaluation [7].

Despite its growing utilization, a notable proportion of MT-sDNA tests are ordered outside guideline-recommended use in average-risk individuals—so-called off-label ordering—which may reduce their effectiveness and increase costs. Off-label ordering includes patients with 1 or more of the following exclusion criteria: personal history of confirmed adenomas, family history of CRC, normal colonoscopy within the preceding 10 years, age greater than 75 years at time of test, prior pelvic radiation exposure, or personal history of colon cancer. Existing studies have demonstrated an MT-sDNA sensitivity of approximately 92% for CRC and 42% for advanced precancerous lesions, with a specificity of 87% in average-risk populations [2]. Community-based data suggest follow-up colonoscopy completion rates of 57-77% after a positive result, and rates of off-label ordering ranging from 18-22%, depending on the clinical setting [8]. Furthermore, understanding follow-up colonoscopy completion rates and pathological outcomes is essential for evaluating the clinical value of this screening strategy.

The present study reports outcomes specifically among patients with positive MT-sDNA test results, including the rate and findings of follow-up colonoscopy, patterns of off-label test ordering, and factors associated with non-adherence to diagnostic evaluation.

Patients and methods

This retrospective study was conducted at a community hospital in the Mid-Atlantic region of the USA and evaluated outcomes of patients who underwent the MT-sDNA test as a screening modality for CRC. All patients identified with a positive MT-sDNA test between May 2023 and May 2024 were included. Patient demographic data extracted from the electronic medical record included age, sex, race, smoking status, alcohol use status, body mass index (BMI), personal histories of pelvic radiation, inflammatory bowel disease (IBD), colon cancer, and family history of colon cancer, the provider type who ordered the MT-sDNA test (i.e., MD/DO vs. Advanced Practice Provider [APP]), and endoscopic and pathological findings after positive MT-sDNA results.

The primary outcome was the proportion of positive MT-sDNA tests among patients who had a follow-up colonoscopy with pathological results, stratified by advanced neoplasia, defined as advanced adenomas (tubular adenoma ≥10 mm, villous or tubulovillous adenoma, serrated adenoma ≥10 mm, high-grade dysplasia) or CRC. Colonoscopy with histopathological evaluation of resected specimens served as the reference standard for the determination of true disease status. Secondary outcomes included assessment of associations between inappropriate MT-sDNA test ordering and: (1) patient demographic characteristics, including age, sex, race, BMI, smoking history and alcohol use history; and (2) provider type. Rates of loss to follow-up for diagnostic colonoscopy were also documented. Pathological outcomes from completed colonoscopies were tabulated on a per-patient basis using frequencies, including tubular adenomas, tubular adenomas >10 mm, villous adenomas, serrated adenomas >10 mm, high-grade dysplasia and CRC.

The WIRB-Copernicus Group deemed this study exempt from institutional review board review under 45 CFR § 46.104(d)(4), as the information recorded by the investigator cannot readily be linked to the subjects and the investigator will not re-identify subjects.

Statistical analysis

Where indicated, descriptive statistics were performed on study variables. Continuous variables, including age, were summarized using means, standard deviations and ranges. Categorical variables were summarized using frequencies and percentages. Statistical analyses of categorical variables were performed using Pearson chi-square tests or likelihood ratio chi-square tests, as appropriate. Independent 2-sample *t*-tests were used to evaluate continuous variables. Analyses comparing racial subgroups were limited to Black and White patients, given the low cell counts in other racial categories. Statistical significance was defined as a 2-sided P-value <0.05. All statistical tests were conducted on de-identified data using Minitab statistical software (Minitab® LLC, 2026).

Results

Between May 2, 2023, and May 2, 2024, a total of 4669 patients had the MT-sDNA test ordered within our electronic medical record system. Of these, 3536 patients completed testing, as described in Fig. 1. A total of 408 patients with positive results were included in this retrospective analysis. The mean age of the cohort was 65.9±8.7 years, with a range of 46-84 years. Baseline characteristics are described in Table 1.

MT-sDNA testing was ordered off-label in 27.2% of patients (n=111). Patients receiving off-label testing were significantly older than those for whom MT-sDNA was ordered in accordance with guideline-supported indications (mean age 70.5 vs. 64.2 years; P<0.001). Among off-label orders, the most frequently met criteria included a family history of CRC (n=40), a normal colonoscopy within the preceding 10 years (n=29), and age greater than 75 years (n=25). A personal history of confirmed adenomas was identified in 22 patients, while prior pelvic radiation was documented in 2 patients, as

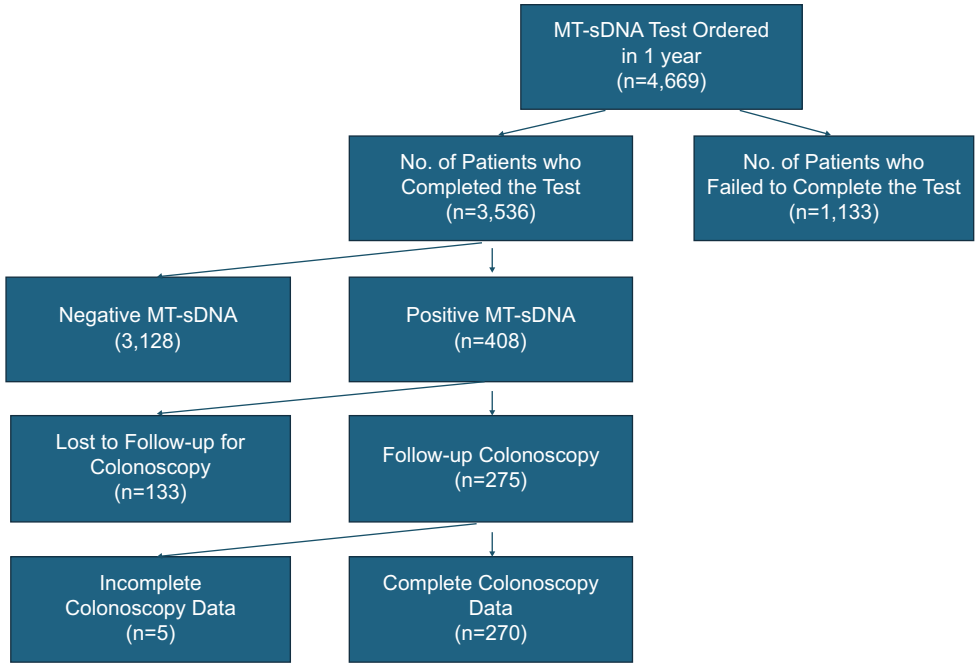

Figure 1 Patients included in the study
MT-sDNA, multitarget stool DNA test
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Table 1 Baseline characteristics of the study population

Characteristic	Value
Age, mean ± SD	65.88±8.74
Age range	46-84
Female, n (%)	233 (57%)
Male, n (%)	175 (43%)
Race, n (%)	
- White	358 (88%)
- Black	42 (10%)
- Hispanic	2 (<1%)
- Other	3 (<1%)
- NA/Declined	3 (<1%)
Smoking status, n (%)	
- Active smoker	89 (22%)
- Former smoker	156 (38%)
- Never smoked	159 (39%)
- NA	4 (1%)
Alcohol use, n (%)	
- Active drinker	215 (53%)
- Former drinker	89 (22%)
- Never drank	99 (24%)
- NA	5 (1%)
BMI category (kg/m^2), n (%)	
- <25	77 (19%)
- 25-30	120 (29%)
- 30-40	181 (44%)
- >40	27 (7%)
- NA	3 (1%)

SD, standard deviation; NA, not available; BMI, body mass index

summarized in Table 2. Some patients fell into more than 1 of these categories.

Additionally, smoking status was significantly associated with incorrect MT-sDNA ordering, with a higher proportion of inappropriate orders observed among patients with a history of smoking (P=0.022). No significant associations were observed between incorrect MT-sDNA ordering and race, sex, alcohol use or BMI; these associations were based on univariate analyses and should be interpreted with caution, as confounding variables were not adjusted for. P-values are presented in Table 3. We also assessed whether there was an association between provider type (MD/DO vs. APP) and MT-sDNA ordering. The analysis showed a significant difference in MT-sDNA ordered incorrectly (P=0.001), with a higher proportion of incorrect orders from MD/DO.

Colonoscopy was clinically indicated for all 408 patients following a positive MT-sDNA result, per current guideline recommendations. Of the 408 patients, 275 (67.4%) underwent colonoscopy. Among those who completed colonoscopy, 58.5% were female and 41.5% were male. Of the 275 patients who underwent colonoscopy, complete pathology data were available for 270; the remaining 5 (1.8%) had incomplete procedural documentation in the electronic medical record and were excluded from the pathology analysis. These missing data appeared to be randomly distributed, with no identifiable pattern related to age, sex or other clinical characteristics. Normal colonoscopy findings were observed in 65 patients (23.6%), while lesions were identified in 205 patients (74.5%); however, the majority of these represented low-risk findings, such as

Table 2 Criterion met for each off-label Cologuard order

Criteria met*	Off-label orders n=111 (%)
Personal history of confirmed adenomas	22
Family history of CRC	40
Normal colonoscopy within last 10 years	29
Age >75 years	25
Pelvic radiation	2
Personal history of colon cancer	0

*: Some patients met more than 1 criterion
CRC, colorectal cancer

Table 3 Cologuard ordered incorrectly by demographics

Demographic Variable	P-Value
Sex	0.417
Race	0.328
Smoking status	0.022
Alcohol use	0.864
BMI	0.176
Age	<0.001
Provider type	0.001

BMI, body mass index

small tubular adenomas, whereas advanced neoplasia, defined as advanced adenomas or colorectal cancer, was identified in only 36 patients (13%). As some patients harbored more than 1 lesion type, the total number of pathological findings recorded exceeds the number of patients with lesions. Table 4 presents findings by lesion type rather than by patient count.

Notably, 133 patients (32.6%) did not undergo follow-up colonoscopy after a positive MT-sDNA result, representing a clinically significant gap in the screening cascade. The mean age of patients who did not complete colonoscopy was 66.9±8.9 years. Among these patients, 72 (54.1%) were female, and 61 (45.9%) were male. There were no significant differences in race or BMI between patients who completed colonoscopy and those who did not. A history of smoking or active tobacco use was present in 80 patients (60.2%) who failed to complete follow-up colonoscopy. Similarly, 93 patients (69.9%) with a history of alcohol use or active drinking did not undergo follow-up colonoscopy.

Among the 270 patients with complete colonoscopy data, 1 patient (0.4%) had CRC, 35 (13.0%) had advanced adenomas, and 140 patients had tubular adenomas, with 61 of these lesions measuring greater than 10 mm. Villous adenomas were identified in 2 patients; none measured more than 10 mm. Serrated adenomas larger than 1 cm were found in 9 patients. Tubulovillous adenomas were seen in 16 patients and high-grade dysplasia was noted in 4 patients. Pathology findings are presented in Table 4.

Table 4 Colonoscopy pathology findings

Finding	n
Tubular adenoma	140
Tubular adenoma >10 mm	61
Villous adenoma	2
Serrated adenoma >10 mm	9
High-grade dysplasia	4
Tubulovillous adenoma	16
Colorectal cancer	1

*: Some patients had more than 1 lesion type identified; values represent number of lesions, not number of patients

Discussion

In our large rural healthcare system, only 11.5% of MT-sDNA tests were positive. The present analysis focuses on outcomes among these positive-test patients and does not represent a full assessment of MT-sDNA screening performance across the tested population. While most patients with a positive MT-sDNA test had at least 1 lesion on follow-up colonoscopy, only 13.3% had advanced adenomas or colorectal cancer, the primary target lesions of MT-sDNA screening [2]. Accordingly, the proportion of positive MT-sDNA tests not associated with advanced neoplasia was 86.7% (diagnostic yield for advanced neoplasia: 13.3%), which is higher than that reported in a prior community-based study (73%) [9]. Among all 3536 patients who completed MT-sDNA testing, colorectal cancer was ultimately identified in 1 (<0.1%), reflecting a population-level detection rate across the full screened cohort; among the 270 patients who completed follow-up colonoscopy with available pathology data, the CRC rate was 1 of 270 (0.4%), reflecting the yield among those who tested positive and underwent diagnostic evaluation. This finding should be interpreted in the context of the study design, which captured only patients with positive MT-sDNA results who subsequently underwent colonoscopy, and does not permit conclusions regarding the overall sensitivity or cancer detection rate of MT-sDNA testing in this population.

Women are consistently shown to participate more frequently in CRC screening compared to men [10]. This disparity is notable, given that males carry a higher risk of CRC incidence and mortality, yet their participation in screening remains suboptimal. In the pivotal large cohort study of the MT-sDNA test, 46.3% of participants were male; at our institution, this trend was mirrored, with 42.9% of screened individuals being male. These findings underscore the need for targeted strategies to improve male engagement in CRC screening, where enhanced counseling by providers and clear communication of risk-benefit considerations may play a pivotal role in narrowing this gap [11].

African American/Black individuals constitute 17.3% of our peninsula population; however, only 10.3% of our study participants were from these groups [12]. In a large cohort reporting MT-sDNA test outcomes, African American

participation was similarly limited at 10.7% [2]. These findings highlight persistent disparities in CRC screening, driven by socioeconomic barriers and inequitable access to healthcare. Addressing these gaps will require multifaceted strategies, including education, community outreach and culturally tailored approaches, to improve minority participation in CRC screening and ensure more equitable preventive care [13].

A substantial proportion of our cohort reported tobacco and alcohol use, both well-established contributors to CRC risk, highlighting the importance of integrating preventive counseling into screening programs. Evidence suggests that smoking cessation and reduction or elimination of alcohol intake could significantly lower the incidence of CRC, underscoring the importance of integrating preventive counseling into screening programs [14]. Addressing modifiable risk factors alongside colorectal cancer screening may enhance the overall effectiveness of prevention strategies, as prior evidence demonstrates that targeted provider education, reminders, and patient-centered interventions significantly improve the completion of diagnostic colonoscopy after positive stool-based tests [15].

The MT-sDNA test was developed to screen average-risk populations. In contrast, colonoscopy remains the preferred modality for individuals at high risk, such as those with a personal or family history of CRC, sessile serrated polyps, or IBD [2]. In our study, 27.2% of MT-sDNA tests were ordered inappropriately, most commonly in patients with a family history of colon cancer, followed by those having a normal colonoscopy in the past 10 years and a prior history of confirmed adenomas. Importantly, MD/DO providers accounted for a disproportionately high share of incorrect MT-sDNA (Cologuard) test orders ($P<0.001$), suggesting an association between provider type and off-label ordering, though this finding requires confirmation in multivariable analyses accounting for practice setting and patient complexity. On univariate analysis, older age and smoking status were associated with inappropriate MT-sDNA ordering; however, these findings should be interpreted cautiously, given the absence of multivariate adjustment for potential confounders. Similar patterns of off-label MT-sDNA ordering, most commonly among patients with a prior normal colonoscopy or a family history of CRC, have been reported in a large community-based cohort; however, the rate of inappropriate ordering in our study was higher (27.2% vs. 20%) [8]. Both the findings of Rao *et al* and our analysis emphasize the need for checks and balances in ordering practices to prevent unnecessary testing, which can lead to false-positive results, patient anxiety, unwarranted invasive procedures and added costs to the health system. Healthcare systems should therefore establish clear institutional policies and feedback mechanisms to minimize inappropriate CRC screening and optimize resource utilization [10].

In our cohort, 32.6% (n=133) of patients with positive MT-sDNA test results did not undergo follow-up colonoscopy. This represents a critical gap in the screening cascade, as a positive MT-sDNA result without subsequent colonoscopy provides no clinical benefit and may create a false sense of security for both patients and providers. Failure to complete diagnostic follow up effectively negates the purpose of stool-based screening and may delay the diagnosis of significant colorectal neoplasia [16]. Although adherence to follow-up colonoscopy is generally higher among MT-sDNA test-positive patients compared to those with positive fecal immunochemical test results (77% vs. 45%), real-world data demonstrate variability, with follow-up colonoscopy rates ranging from 57% to 77% depending on demographic factors [9,17]. Among patients who did not undergo diagnostic colonoscopy, 60.2% were active smokers or had a history of tobacco use, and 69.9% reported current or prior alcohol consumption. These findings suggest an association between substance use and failure to complete diagnostic follow up, though causal inference is limited by the absence of multivariable adjustment for socioeconomic, logistical and clinical confounders. Targeted identification and education of higher-risk populations, including individuals with a history of smoking or alcohol use, may represent an important strategy to improve adherence to follow-up colonoscopy after positive stool-based screening [16].

Several limitations of this study warrant consideration. First, the single-center, retrospective design limits its generalizability to institutions with different patient demographics, practice patterns or resource availability. Second, selection bias is an inherent concern, as the cohort was restricted to patients with positive MT-sDNA results who were captured within a single electronic medical record system; patients who were lost to follow up prior to test completion, or who sought care outside our system, were not captured. Third, 32.6% of patients did not complete follow-up colonoscopy, introducing the possibility that significant neoplasias were missed in this subgroup, and that our estimates of diagnostic yield are subject to ascertainment bias. Fourth, all associations reported, including those involving smoking status, alcohol use and provider type, are based on univariate analyses and were not adjusted for potential confounders; these should therefore be considered hypothesis-generating rather than definitive. Future prospective, multicenter studies incorporating multivariable adjustment will be necessary to validate these associations and better characterize MT-sDNA utilization patterns across diverse healthcare settings.

In conclusion, our rural, community-based analysis revealed a high rate of off-label multitarget stool DNA test utilization, accompanied by a substantial proportion of positive tests not associated with advanced neoplasia (advanced adenomas or colorectal cancer). Notably, incomplete completion of diagnostic colonoscopy following a positive MT-sDNA test was more common among patients with documented tobacco use and alcohol use disorders. While causal inferences cannot be drawn from this observational design, our results support the potential value of targeted provider education and patient-centered interventions to promote guideline-concordant test ordering and improve follow-up colonoscopy completion rates. Prospective, multicenter studies are needed to confirm these associations and evaluate the impact of such interventions on screening outcomes and resource utilization.

Summary Box

What is already known:

- Multitarget stool DNA (MT-sDNA; Cologuard®) testing is an FDA-approved, noninvasive colorectal cancer (CRC) screening test recommended for average-risk individuals, with high sensitivity for CRC and advanced adenomas

What the new findings are:

- In a large, rural, community-based healthcare system, more than one-quarter (27%) of MT-sDNA tests were ordered off-label, most commonly for patients with a family history of CRC, a recent normal colonoscopy, or advanced age
- Furthermore, 33% of patients failed to complete the follow-up colonoscopy, even after a positive MT-sDNA test

References

1. Patel SG, Karlitz JJ, Yen T, Lieu CH, Boland CR. The rising tide of early-onset colorectal cancer: a comprehensive review of epidemiology, clinical features, biology, risk factors, prevention, and early detection. *Lancet Gastroenterol Hepatol* 2022;**7**:262-274.
2. Imperiale TF, Ransohoff DF, Itzkowitz SH, et al. Multitarget stool DNA testing for colorectal-cancer screening. *N Engl J Med* 2014;**370**:1287-1297.
3. Brenner H, Chen C. The colorectal cancer epidemic: challenges and opportunities for primary, secondary and tertiary prevention. *Br J Cancer* 2018;**119**:785-792.
4. Siegel RL, Fedewa SA, Anderson WF, et al. Colorectal cancer incidence patterns in the United States, 1974-2013. *J Natl Cancer Inst* 2017;**109**:djw322.
5. Kleinschmidt TK, Clements A, Parker MA, Scarcliff SD. Retrospective review of multitarget stool DNA as a screening test for colorectal cancer. *Am Surg* 2023;**89**:603-606.
6. Patel M, Bierle-Karnes L, Hasan S, Lavette LE, Copland AP. S349 appropriateness of Cologuard testing at an academic medical center. *Am J Gastroenterol* 2023;**118**:S282.
7. Lazar NJ, Khreisat A, AlKloub R, Al-Hillan A, Duffy M. Inappropriate multi-target stool DNA use for colorectal cancer screening: risks, compliance, and outcomes. *Cureus* 2023;**15**:e40506.
8. Ahn AB, Kulhari S, Rhee J, Cooper G. Inappropriate ordering of multitarget stool DNA tests for colon cancer screening. *J Am Board Fam Med* 2024;**37**:328-331.
9. Rao AK, Kalra S, Tran D, et al. The utility of multitarget stool DNA testing in community-based clinical practice. *Am J Gastroenterol* 2026;**121**:441-445.
10. Pallesen AVJ, Herrstedt J, Westendorp RGJ, Mortensen LH, Kristiansen M. Differential effects of colorectal cancer screening across sociodemographic groups in Denmark: a register-based study. *Acta Oncol* 2021;**60**:323-332.
11. Shaukat A, Kahi CJ, Burke CA, Rabeneck L, Sauer BG, Rex DK. ACG clinical guidelines: colorectal cancer screening 2021. *Am J Gastroenterol* 2021;**116**:458-479.
12. Eastern Shore Regional GIS Cooperative. Mapping Census 2020 on Delmarva. ArcGIS StoryMaps [Internet]. 2022. Available from: https://storymaps.arcgis.com/stories/e71ae447edec475aa4cc8076788e25c5 [Accessed 27 August 2026].
13. Rutter CM, Knudsen AB, Lin JS, Bouskill KE. Black and white differences in colorectal cancer screening and screening outcomes: a narrative review. *Cancer Epidemiol Biomarkers Prev* 2021;**30**:3-12.
14. Siegel RL, Kratzer TB, Giaquinto AN, Sung H, Jemal A. Cancer statistics, 2025. *CA Cancer J Clin* 2025;**75**:10-45.
15. Selby K, Baumgartner C, Levin TR, et al. Interventions to improve follow-up of positive results on fecal blood tests: a systematic review. *Ann Intern Med* 2017;**167**:565-575.
16. Cusumano VT, Corona E, Partida D, Yang L, Yu C, May FP. Patients without colonoscopic follow-up after abnormal fecal immunochemical tests are often unaware of the abnormal result and report several barriers to colonoscopy. *BMC Gastroenterol* 2020;**20**:115.
17. Greene M, Camardo M, Karlitz JJ, et al. Adherence to multi-target stool DNA test and time to follow-up colonoscopy: real-world analysis of patients from the Beth Israel Lahey Health. *Curr Med Res Opin* 2025;**41**:1811-1823.